Patent claims

1. Recombinant microbial cell comprising

i) a first enzyme activity controlling assimilation of a nitrogen nutrient source,

wherein the first enzyme activity is encoded by a first nucleic acid operable linked to an expression signal not natively associated with the first nucleic acid, and

wherein the expression of the first enzyme activity is increased as compared to the expression of the first enzyme activity when the first nucleic acid is associated with its native expression signal,

and/or

ii) a second enzyme activity controlling assimilation of a nitrogen nutrient source,

wherein the second enzyme activity is encoded by a second nucleic acid operable linked to an expression signal not natively associated with the second nucleic acid, and

wherein the expression of the second enzyme activity is increased as compared to the expression of the second enzyme activity when the second nucleic acid is associated with its native expression signal,

the cell further comprising

iii) a reduced or eliminated expression of a third enzyme activity encoded by a third nucleic acid and controlling assimilation in the cell of a nitrogen nutrient source,

wherein the expression of the third enzyme activity is reduced or eliminated as compared to the expression of the third enzyme activity when the third nucleic acid is associated with its native expression signal.

2. Microbial cell according to claim 1, the cell comprising

i) a further enzyme activity, the further enzyme activity mediates an energy yielding first reaction resulting in a production of a first metabolite, wherein

ii) the first reaction being operably linked to an energy requiring second reaction resulting in assimilation of a nutrient source.

3. Microbial cell according to claim 2, wherein the energy requiring second reaction resulting in assimilation of a nutrient source is controlled at least by the first and/or second enzyme activity.

4. Microbial cell according to claim 1, the cell comprising

i) a further enzyme activity, wherein the further enzyme activity mediates an energy yielding first reaction resulting in a production of a first metabolite, wherein

ii) the further enzyme activity, when expressed at an increased level, results in an increased production of the first metabolite, wherein

iii) the increased expression of the further enzyme activity and/or the increased production of the first metabolite is operably linked to an increased expression of the first and/or second enzyme activity.

5. Microbial cell according to claim 1, the cell being selected from the group consisting of a fungal cell, a yeast cell, and a bacterial cell.

6. Microbial cell according to claim 6, the cell being a yeast cell.

7. Microbial cell according to claim 1, wherein the nitrogen source is ammonia and/or an ammonium ion.

8. Microbial cell according to claim 1 wherein the first or second enzyme activity is mediating at least one enzymatic reaction resulting in the assimilation of ammonia and/or an ammonium ion in the microbial cell.

9. Microbial cell according to claim 1 wherein the first and second enzyme activities are each mediating at least one enzymatic reaction resulting in the assimilation of ammonia in the microbial cell.

10. Microbial cell according to claim 1 wherein at least one of the first and second enzyme activities is mediating a biosynthetic reaction.

11. Microbial cell according to claim 1 wherein the first enzyme activity is a glutamate synthase activity.

12. Microbial cell according to claim 11 wherein the activity is a *Saccharomyces cerevisiae* glutamate synthase, or a functionally equivalent activity capable of catalysing a glutamate synthase reaction.

13. Microbial cell according to claim 12 wherein the activity is that encoded by *GLT1* of *Saccharomyces cerevisiae* as deposited under DSM Accession Number 12275.

14. Microbial cell according to claim 1 wherein the second enzyme activity is a glutamine synthetase activity.

15. Microbial cell according to claim 14 wherein the activity is a *Saccharomyces cerevisiae* glutamine synthetase activity, or a functionally equivalent activity capable of catalysing a glutamine synthetase reaction.

16. Microbial cell according to claim 15 wherein the activity is that encoded by *GLN1* of *Saccharomyces cerevisiae* as deposited under DSM Accession Number 12274.

17. Microbial cell according to claim 1 wherein the third enzyme activity is a glutamate dehydrogenase activity.

18. Microbial cell according to claim 17 wherein the activity is a *Saccharomyces cerevisiae* glutamate dehydrogenase activity, or a functionally equivalent activity capable of catalysing a glutamate dehydrogenase reaction.

19. Microbial cell according to claim 18 wherein the activity is that encoded by *GDH1* of *Saccharomyces cerevisiae*.

20. Microbial cell according to claim 1 wherein the expression of the third enzyme activity is reduced by at least 50%.

21. Microbial cell according to claim 1 wherein the third enzyme activity is not expressed.

22. Microbial cell according to claim 1 wherein the third enzyme activity has been eliminated.

23. Microbial cell according to claim 21 or 22 wherein a nucleotide sequence encoding the third enzyme activity and/or an expression signal directing expression of the activity has been partly or wholly deleted from a chromosomal replicon and/or an extrachromosomal replicon harboured by the microbial cell.

24. Microbial cell according to any of claims 20 to 23 wherein the third enzyme activity is a glutamate dehydrogenase activity.

25. Microbial cell according to claim 24 wherein the glutamate dehydrogenase activity is that encoded by *GDH1* of *Saccharomyces cerevisiae*, or a functionally equivalent activity capable of catalysing a glutamate dehydrogenase reaction.

26. Microbial cell according to claim 25, the cell being *Saccharomyces cerevisiae* TN19 as deposited under Accession Number DSM 12276.

27. Microbial cell according to claim 25, the cell being *Saccharomyces cerevisiae* TN22 as deposited under Accession Number DSM 12277.

28. Microbial cell according to any of claims 20 to 27 wherein the expression of the first or second enzyme activity is increased by a factor of at least 1.5.

29. Microbial cell according to any of claims 20 to 28 wherein the expression of the first and second enzyme activities are increased by a factor of at least 1.5.

30. Microbial cell according to any of the previous claims and further comprising a fourth nucleic acid encoding a fourth enzyme activity controlling an intracellular redox system of the cell, wherein the fourth nucleic acid is operably linked to an expression signal not natively associated with the fourth nucleic acid.

31. Microbial cell according to claim 30 wherein the fourth enzyme activity is an intracellular transhydrogenase activity.

32. Microbial cell according to claim 30 wherein the transhydrogenase activity is that encoded by *CTH* of *Azotobacter vinelandii* as harboured by *Saccharomyces cerevisiae* TN4 deposited under DSM Accession Number 12267.

33. Microbial cell according to any of the previous claims in the form of a frozen or freeze-dried preparation such as a lyophilisate.

34. Composition comprising the microbial cell according to any of the previous claims and a carrier.

35. Composition according to claim 34 wherein the carrier is a physiologically acceptable carrier such as a water-based liquid, preferably a broth suitable for culturing the microbial cell.

36. Composition according to claim 34 or 35, wherein the composition is a fermentation starter culture.

37. Composition according to any of claims 34 to 36 for use in the production of a first metabolite.

38. Microbial cell according to any of claims 1 to 33 for use in the production of a first metabolite.

39. Microbial cell according to claim 38, wherein the increased expression of the first and/or second enzyme activity encoded by the first and/or second nucleic acid, respectively, results in an increased production of a first metabolite, the production being increased as compared to the production of the metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

40. Microbial cell according to claim 39, wherein the production of the first metabolite is increased by a factor of at least 1.10.

41. Microbial cell according to claim 38 wherein the microbial cell is a yeast cell.

42. Microbial cell according to claim 38 wherein the first metabolite is ethanol.

43. Microbial cell according to claim 41, wherein the yeast cell further produces a second metabolite, the production of the second metabolite being decreased as compared to the production of the metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

44. Microbial cell according to claim 43 wherein the second metabolite is glycerol.

45. Microbial cell according to any of claims 1 to 33 and 38 to 44, or the composition according to any of claims 34 to 37, for use in a preparation of a drinkable or an edible product.

46. Microbial cell according to any of claims 1 to 33 and 38 to 44, or the composition according to any of claims 34 to 37, for use in a production of a first metabolite for use in a drinkable or an edible product.

47. Microbial cell according to claim 46 wherein the first metabolite provides a desirable organoleptic quality to the product.

48. Microbial cell according to claim 46, wherein the metabolite is ethanol.

49. Use of the microbial cell according to any of claims 1 to 33 and 38 to 44, or the composition according to any of claims 34 to 37, in a production of a first metabolite.

50. Use of claim 49, wherein the production of the first metabolite is increased in a cell wherein the expression of the first and/or second enzyme activity encoded by the first and/or second nucleic acid, respectively, is increased as compared to the production of the first metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

51. Use of claim 49, wherein the microbial cell is a yeast cell.

52. Use of claim 49, wherein the first metabolite is ethanol.

53. Use according to claim 51 or 52, wherein the yeast cell further produces a second metabolite, the production of the second metabolite is decreased as compared to the production of the second metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

54. Use according to claim 53 wherein the second metabolite is glycerol.

55. Use according to claim 54 in the preparation of a drinkable or an edible product.

56. Method of producing a first metabolite, the method comprising the steps of

i) cultivating the microbial cell according to any of claims 1 to 33 and 38 to 44, or the composition according to any of claims 34 to 37, in a suitable growth medium and under such conditions that the microbial cell is producing a first metabolite, and optionally

ii) isolating the first metabolite in a suitable form, and further optionally

iii) purifying the isolated first metabolite.

57. Method of claim 56, wherein the production of the first metabolite is increased in a cell wherein the expression of the first and/or second enzyme activity encoded by the first and/or second nucleic acid, respectively, is increased, as compared to the production of the first metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

58. Method of claim 57, wherein the production of the metabolite is increased by a factor of at least 1.02, such as 1.04, for example 1.08, such as 1.16, for example 1.25, such as 1.4.

59. Method of claim 56 wherein the microbial cell is a yeast cell.

60. Method of claim 56 wherein the first metabolite is ethanol.

61. Method of claim 56, wherein the yeast cell further produces a second metabolite, the production of the second metabolite is decreased as compared to the production of the second metabolite in a cell wherein the first and/or second nucleic acid is associated with a native expression signal.

62. Method of claim 61 wherein the second metabolite is glycerol.

63. Method of constructing a microbial cell according to any of claims 1 to 33, and 38 to 48, the method comprising the steps of

i) operably linking a nucleotide sequence encoding the first enzyme activity with an expression signal not natively associated with the first nucleotide sequence, wherein the expression of the first enzyme activity is increased as compared to the expression of the first enzyme activity when the first nucleic acid is associated with its native expression signal, and/or

ii) operably linking a nucleotide sequence encoding the second enzyme activity with an expression signal not natively associated with the second nucleotide sequence, wherein the expression of the second enzyme activity is increased as compared to the expression of the second enzyme activity when the second nucleic acid is associated with its native expression signal, and

iii) eliminating the third enzyme activity from the microbial cell or operably linking a nucleotide sequence encoding the third enzyme activity with an expression signal not natively associated with the nucleotide sequence, wherein the expression of the third enzyme activity is decreased as compared to the expression of the third enzyme activity when the third nucleic acid is associated with its native expression signal, and

iv) introducing the operably linked nucleotide sequences obtained under steps i) and/or ii) and iii), into the microbial cell, or

v) introducing the operably linked nucleotide sequences obtained under steps i) and/or ii), into a microbial cell wherein the third enzyme activity has been eliminated.

64. Method of claim 63, wherein the microbial cell is selected from the group consisting of a fungal cell, a yeast cell, and a bacterial cell.

65. Method of claim 64, wherein the microbial cell is a yeast cell.

66. Method of claim 64 wherein the first enzyme activity is a glutamate synthase activity.

67. Method of claim 66, wherein the glutamate synthase activity is that encoded by *GLT1* of *Saccharomyces cerevisiae* TN17 as deposited under DSM Accession Number 12275, or a functionally equivalent activity capable of catalysing a glutamate synthase reaction.

68. Method of claim 64 wherein the second enzyme activity is a glutamine synthetase activity.

69. Method of claim 68 wherein the glutamine synthetase activity is that encoded by *GLN1* of *Saccharomyces cerevisiae* TN15 as deposited under DSM Accession Number 12274, or a functionally equivalent activity capable of catalysing a glutamine synthetase reaction.

70. Method of claim 64 wherein the third enzyme activity, when present in the microbial cell, is a glutamate dehydrogenase activity, preferably one encoded by *GDH1* of *Saccharomyces cerevisiae*.

71. Method of claim 64 wherein the cell is *Saccharomyces cerevisiae* TN19 as deposited under DSM Accession Number 12276.

72. Method of any of claims 63 to 71, the method comprising a further step of freezing or freeze-drying the microbial cell in the preparation of a reconstitutable lyophilisate.